REMARKS

In an Office Action mailed December 31, 2007, the Examiner in charge of the application maintained a single rejection of pending claims 1, 3, 7, 9, 10, 12, 13, 15, and 16 under 35 U.S.C. § 103(a) as being unpatentable over Priori *et al.* (1996) in view of Gepstein *et al.* (evidenced by US provisional application 60/306,462).

Applicants thank the undersigned for his time during a personal interview at the US Patent and Trademark Office on February 15, 2008. The Interview Summary mailed on March 4, 2008 accurately summarizes the substance of the Interview.

Rejections Under 35 U.S.C. § 103

The Examiner maintains that the invention as claimed would be obvious under §103(a) over Priori in view of Gepstein. The application of Priori to the rejected claims is not at issue. In particular, however, the Examiner maintains that the method steps in Applicant's claims for deriving cardiomyocytes from human embryonic stem cells (hESCs) are not patentably distinct from the disclosure of Gepstein.

Applicants amend the independent claims to recite a point of difference from Gepstein that, in the view of a skilled artisan, would (at the very least) affect differentiation of cardiomyocytes from hESC-derived embryoid bodies (EBs). More to the point, however, the differences between the Gepstein disclosure and Applicants' methods as now claimed are sufficient to preclude a skilled person from concluding that cardiomyocyte-containing cultures of Gepstein would have had the recited properties of the claims. Most pertinently, the added culturing step finds no support or basis in Gepstein.

The amended claims now recite a culturing step for obtaining embryoid bodies from hESCs. In particular, the claims recite that aggregates containing 500-800 undifferentiated hESCs are cultured to produce EBs that are subsequently differentiated for between 40 and 95 days to yield the indicated cardiomyocyte cell types. Support for this amendment is found in the application as filed at paragraph [00027], which states that "ES cell colonies were dispersed into cell aggregates containing approximately 500-800 cells using 1 mg/ml dispase. The cell

aggregates were then cultured in suspension in cell culture flasks (BD Bioscience) with ES cell medium without basic fibroblast growth factor for 6 days with media changed daily. To promote cardiac differentiation, 6-day old EBs were transferred to the 6-well plates coated with 0.1% gelatin in media consisting of DMEM supplemented with 15% FBS (selected for cardiac differentiation, 2 mmol/L L-glutamine, and 1% nonessential amino acids." The same paragraph describes obtaining beating EBs.

In contrast, Gepstein indicated that "To induce differentiation, ES cells were dispersed to small clamps (sic, clumps) (3-20 cells) using collagenase IV (Life Technologies, Inc., 1 mg/ml for 20 min). The cells were transferred to plastic Petri dishes (Miniplast, Ein Shemer, Israel), at a cell density of about 5 x 10^6 cells in a 58-mm dish, where they were cultured in suspension for 7-10 d." Provisional application, page 3 ("Methods"), second paragraph (ES cell preparation and production of EBs). After the two manuscripts and before the figures, Gepstein's provisional application included a list of "Applications relevant to the present invention" (pages 36-38) and a set of three claims. In discussing on page 36 the generation of cardiomyocytes and [their] isolation, Gepstein stated that " Our experiment demonstrated that the optimal timing is 7-10 days for growth in suspension before plating."

Applicants have identified a clear and ascertainable difference in starting material compared to Gepstein's starting material. The foregoing confirms that Applicants' hESC aggregates and Gepstein's hESC clumps are maintained in distinct microenvironments both before, during and after suspension culture. Especially early in the suspension culture, the sheer difference in aggregate/clump cell number translates into differences in cell-cell contacts and in the available concentrations of secreted factors. Likewise, the time spent in suspension culture is also different. While the skilled artisan would from these differences not have been able to predict the precise cell profile of either Applicants' or Gepstein's beating EBs, the differences in starting materials and culture conditions (not to mention the previously argued differences in subsequent culture times, incorporated by reference herein), would have provided the skilled artisan with a reasonable expectation of differences between the resulting cultures. Applicants

do not recite the differences in suspension culture time in the claims and do not rely upon this difference as a basis for patentability; Applicants simply point out that as of either Applicants' or Gepstein's filing date, a skilled person would have understood that by the time the cells of the two cultures had differentiated into cardiomyocytes, they would have been exposed to different microenvironments, different nearby cell types, and different cell growth factors for different lengths of time. Just as one would expect real differences between human adults raised in different cultures, near different people, eating different foods, one would likewise expect differences in the beating EB products of the two methods.

Although the Examiner maintained his demand for evidence that Applicants' culture is patentably distinct from Gepstein's, Applicants have made a diligent effort to articulate clear and direct reasons why the skilled person would have had no reason to presume that the two would be the same, and indeed why the more reasoned view would expect the two to be different. If the Examiner is still inclined to believe that Applicants have not overcome the rejection for obviousness under §103(a), Applicants remind the Examiner that the claims at issue here recite a method step (culturing aggregates of approximately 500 to 800 undifferentiated human embryonic stem cells) not disclosed or contemplated by Gepstein. This difference in cell number is not a mere few percent, from which a skilled person could, *arguendo*, readily adjust the culture conditions. At the closest point between the two disclosures, an approximately 500 cell aggregate in Applicants' method contains about 25 times more cells than the 20 cells of Gepstein's clumps. Indeed, in some embodiments, Applicants' aggregates would contain more than 250 times as many cells as Gepstein's. In view of Gepstein's clear teaching of a small clump size, Gepstein cannot be said to disclose, teach or suggest Applicants' culturing step. Moreover, the role in culturing hESCs of cell number, cell microenvironment and cell contacts underscores that the Gepstein cultures would not have had the properties of the claimed invention.

Reconsideration is respectfully requested.

Fees

A petition for an extension of time for three months accompanies this response so the response will be deemed to have been timely filed. Should any additional extension of time be due, in this or in any subsequent response, please consider this to be a petition for the appropriate extension of time and a request to charge the petition fee to deposit account number 17-0055. No other fee is believed due. However, should any be due in this or any subsequent response, please consider this a request to charge the fee to the same deposit account.

Respectfully submitted,

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